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(54) Title: COMPOSTIONS FOR DARKENING THE SKIN

(57) Abstract: The present invention relates to the use of He Shou Wu extract in darkening the skin.

COMPOSITIONS FOR DARKENING THE SKIN

FIELD OF THE INVENTION

The present invention relates to the use of He Shou Wu extract in darkening the skin.

BACKGROUND OF THE INVENTION

The darkening of skin color is a concern for many individuals. Most people obtain darker skin through exposure to UV light (e.g., suntanning or UV lamps). Production of melanin, and the type of melanin when stimulated by UV are genetically determined. UV exposure, however, results in accelerated skin aging and increased incidence of skin cancer. The ability to generate a tanned appearance without incurring photodamage, thus, is important to many individuals. Accordingly, alternative methods for "sunless tanning" have evolved.

One method is the use of products containing dihydroxy acetone (DHA). Some of these products, however, produce color that is too orange and unnatural to the user. Moreover, the DHA-produced skin color only minimally protects the user from UV irradiation. Products containing beta-carotene, cantaxanthin and lycopene have also been used to darken the skin. These products, however, have no effect at all on melanogenesis and usually result in unnatural and uneven distributed skin color by saturating and staining the fat layers just below the skin. In addition, these products do not provide any sun-protection as compared to naturally tanned skin. Melanotan and MelanX are synthetic hormone drugs that mimic the action of melanocyte-stimulating hormone (MSH) and are used to darken the skin only when administered by injection, not orally or topically.

Psoralens, on the other hand, work by making the skin hypersensitive to the sun and therefore melanin production is accelerated. They do not make the skin darker without exposure to UV, and that exposure must be carefully regulated to minimize the serious risk for skin cancer. Psoralens in conjunction with medical grade UV lamps are an accepted treatment for people afflicted with vitiligo and psoriasis, but are not recommended for patients with fair skins. Thus, a product is desired that could enhance the body's natural pigment content, resulting in a desired skin color and enhanced photo-protection without the need of UV exposure.

He Shou Wu has traditionally believed to be useful in nourishing the kidney and liver, preventing premature graying of the hair, relief of constipation, and skin lesions. It was also used for treating hyperglycemia (PCT Patent Application No. WO 95/30427 and U.S. Patent No. 5,531,991), increasing insulin activity (PCT Patent Application No. WO 99/22752 and U.S. Patent No. 6,200,569) and inhibiting testosterone 5 alpha-reductase. He Shou Wu was traditionally administered in combination with other herbs by oral applications (PCT Patent Application Nos. WO 97/10833 and WO 01/22934). However, it has been found that the activity of He Shou Wu and other herbs is greatly reduced by stomach acids before they have had an opportunity to be absorbed into the blood stream. U.S. Patent No. 5,464,443 and PCT Patent Application No. WO 97/10833 disclose an oral composition for darkening hair color in a toothpaste or chewing gum base

containing He Shou Wu and other herbs through buccal absorption, which greatly increases the effectiveness of the herbs.

A number of prior arts attempt to use He Shou Wu in combination with other herbs for promoting hair growth including preventing or minimizing hair loss by external application and darkening hair (PCT Patent Application No. WO 91/12792).

The present inventors, however, have unexpectedly discovered that He Shou Wu is effective for darkening the skin.

SUMMARY OF THE INVENTION

In one aspect, the present invention relates to a composition for darkening the skin comprising a safe and effective amount of a He Shou Wu extract and a cosmetically acceptable carrier. In another aspect, the present invention relates to a method of darkening the skin comprising topically applying to the skin a composition comprising a safe and effective amount of a He Shou Wu extract. In another aspect, the present invention relates to a product comprising: (a) a composition for darkening the skin, wherein such composition comprises a safe and effective amount of a He Shou Wu extract; and (b) instructions directing the user to apply said composition to the skin to darken the skin.

In still another aspect, the present invention relates to a method of promoting a product comprising a

composition where such composition comprises a safe and effective amount of a He Shou Wu, wherein such method comprises directing the user to apply such composition to the skin to darken the skin.

Other features and advantages of the present invention will be apparent from the detailed description of the invention and from the claims

DETAILED DESCRIPTION OF THE INVENTION

It is believed that one skilled in the art can, based upon the description herein, utilize the present invention to its fullest extent. The following specific embodiments are to be construed as merely illustrative, and not limitative of the remainder of the disclosure in any way whatsoever.

Unless defined otherwise, all technical and scientific terms used herein have the same meaning as commonly understood by one of ordinary skill in the art to which the invention belongs. Also, all publications, patent applications, patents, and other references mentioned herein are incorporated by reference. Unless otherwise indicated, a percentage refers to a percentage by weight (i.e., %(W/W)).

Definitions

What is meant by "darkening the skin" is darkening the appearance of the skin, including, but not limited to, tanning the skin.

What is meant by a "product" is a product in finished packaged form. In one embodiment, the package

is a container such as a plastic, metal or glass tube or jar containing the composition. The product may further contain additional packaging such as a plastic or cardboard box for storing such container. In one embodiment, the product contains instructions directing the user to apply said composition to the skin to darken the skin (e.g., to tan the skin) or even skin tone (e.g., to darken light areas of the skin or to treat or prevent mottled hyperpigmentation). Such instructions may be printed on the container, label insert, or on any additional packaging.

What is meant by "promoting" is promoting, advertising, or marketing. Examples of promoting include, but are not limited to, written, visual, or verbal statements made on the product or in stores, magazines, newspaper, radio, television, internet, and the like. Examples of such statements include, but are not limited to, "evens skin tone," "darkens the skin," "prevents, reduces, or treats mottled hyperpigmentation," "tans the skin," or "sunless tan."

As used herein, "topically applying" means directly laying on or spreading on outer skin, e.g., by use of the hands or an applicator such as a wipe, roller, or spray.

As used herein, "cosmetically-acceptable" means that the ingredients which the term describes are suitable for use in contact with tissues (e.g., the skin) without undue toxicity, incompatibility, instability, irritation, allergic response, and the like.

As used herein, "safe and effective amount" means an amount of the He Shou Wu extract or composition sufficient to induce a darkening of the skin, but low enough to avoid serious side effects. The safe and effective amount of the extract or composition will vary with the area being treated, the age and skin type of the end user, the duration and nature of the treatment, the specific extract or composition employed, the particular cosmetically-acceptable carrier utilized, and like factors.

He Shou Wu Extract

What is meant by a "He Shou Wu extract" is a blend of compounds isolated from the plant Polygonum Multiflorum. In one embodiment, the compounds are isolated from the root of the plant (e.g., radix polygoni multiflori). Such compounds may be isolated from a part(s) of the plant (e.g., the seed, root, rhizome, fruit and/or leaf of the plant) by physically removing a piece of such plant, such as grinding a root of the plant. Such compounds may also be isolated from the plant by using extraction procedures well known in the art (e.g., the use of organic solvents such as lower C_1-C_8 alcohols, C_1-C_8 alkyl polyols, C_1-C_8 alkyl ketones, C_1-C_8 alkyl ethers, acetic acid C_1-C_8 alkyl esters, and chloroform, and/or inorganic solvents such as water, inorganic acids such as hydrochloric acid, and inorganic bases such as sodium hydroxide). In one embodiment, the He Shou Wu extract contains only

hydrophilic compounds (e.g., isolated by using a hydrophilic solvent, such as water or ethanol). In one embodiment, the He Shou Wu extract is an aqueous extract from the root.

The amount of the He Shou Wu extract present in the composition will depend on the type of extract used. The extract typically will be present in the composition in an amount from about 0.001% to about 20% by weight, in particular in an amount from about 0.01% to about 5% by weight.

Pigment

In one embodiment, the composition of the present invention further comprises a pigment. What is meant by a "pigment" is a compound(s) that can be taken up by epidermal cells, resulting in visually darker look to the skin. Examples of such pigments include, but not limiting to, melanin and melanin derivatives (e.g, both melanin polymers and lower molecular weight water-soluble melanin derivatives); extracts from natural sources containing pigments (e.g., brown pigments from plants from the Hedychium genus or Bearberry genus or yellow, orange and red pigments, from plants containing carotenoids or canthaxanthins); or synthetic chemicals such as compounds containing copper (e.g., copper salts such as $CuCl_2$) or synthetic carotenoids or canthaxantins. Examples of synthetic melanin derivatives are disclosed in U.S. Patent Nos. 5,618,519, 5,384,116, and 5,227,459. Examples of

soluble melanin derivatives are disclosed in U.S. Patent Nos. 5,744,125, 5,225,435, 5,218,079, and 5,216,116. Examples of commercially available soluble melanin derivatives include Melasyn-100™ from San-mar laboratories, Inc. (Elmsford, NY) and MelanZe™ from Zylepsis (Ashford, Kent, United Kingdom).

The amount of pigment present in the composition will depend on the type of pigment used. The pigment typically will be present in the composition in an amount from about 0.001% to about 20% by weight, in particular in an amount from about 0.005% to about 5% by weight.

Peptides

In one embodiment, the composition of the present invention further comprises a peptide of the Formula I

R_1

$>A_1\text{-}A_2\text{-}A_3\text{-}A_4\text{-}A_5\text{-}A_6\text{-}A_7\text{-}R_3$ Formula I

R_2

wherein:

A_1 is Ser or 2,3-diaP, or is absent;

A_2 is Val, Leu, Ile, or Cha;

A_3 is Val, Leu, Ile, or Cha;

A_4 is Gly or Ala;

A_5 is Lys, Arg, or Har;

A_6 is Val, Leu, Ile, or Cha, or is absent;

A_7 is Asp or Glu, or is absent; provided, A_7 is absent if A_6 is absent;

each R_1 and R_2, independently, is H, C_{1-12} alkyl, C_{7-10} phenylalkyl, or $C(=O)E_1$, where E_1 is C_{1-20} alkyl, C_{3-20} alkenyl, C_{3-20} alkynyl, phenyl, 3,4-dihydroxyphenylalkyl, naphthyl, or C_{7-10} phenylalkyl; provided that when either R_1 or R_2 is $C(=O)E_1$, the other must be H; and

R_3 is OH, NH_2, C_{1-12} alkoxy, C_{7-10} phenylalkoxy, C_{11-20} naphthylalkoxy, C_{1-12} alkylamino, C_{7-10} phenylalkylamino, or C_{11-20} naphthylalkylamino;

or a cosmetically acceptable salt thereof.

In one embodiment, R_1 and R_2, which are bound to the N-terminus of the peptide, are both H. In another embodiment, R_1 is H and R_2 is $C(=O)E_1$ (e.g., palmitoyl, oleatoyl, or stearatoyl).

Examples of peptides of the present invention include, but are not limited to, to H_2-Leu-Ile-Gly-Arg-NH_2 (Peptide 1, SEQ ID NO:1), H_2-Leu-Ile-Gly-Arg-Leu-NH_2(Peptide 2, SEQ ID NO:2), H_2-Leu-Ile-Gly-Lys-NH_2 (Peptide 3, SEQ ID NO:3), H_2-Ser-Leu-Ile-Gly-Lys-NH_2 (Peptide 4, SEQ ID NO:4), H_2-Leu-Ile-Gly-Arg-OH (SEQ ID NO:5), H_2-Leu-Ile-Gly-Arg-Leu-OH (SEQ ID NO:6), H_2-Leu-Ile-Gly-Lys-OH (SEQ ID NO:7), H_2-Ser-Leu-Ile-Gly-Lys-OH (SEQ ID NO:8), Palmitoyl-Leu-Ile-Gly-Arg-NH_2 (SEQ ID NO:9), Palmitoyl-Leu-Ile-Gly-Arg-Leu-NH_2(SEQ ID NO:10), Palmitoyl-Leu-Ile-Gly-Lys-NH_2 (SEQ ID NO:11), Palmitoyl-Ser-Leu-Ile-Gly-Lys-NH_2 (SEQ ID NO:12), Palmitoyl-Leu-Ile-Gly-Arg-OH (SEQ ID NO:13), Palmitoyl-Leu-Ile-Gly-Arg-Leu-OH (SEQ ID NO:14),

Palmitoyl-Leu-Ile-Gly-Lys-OH (SEQ ID NO:15), Palmitoyl-Ser-Leu-Ile-Gly-Lys-OH (SEQ ID NO:16), Stearatoyl-Leu-Ile-Gly-Arg-NH_2 (SEQ ID NO:17), Stearatoyl-Leu-Ile-Gly-Arg-Leu-NH_2(SEQ ID NO:18), Stearatoyl-Leu-Ile-Gly-Lys-NH_2 (SEQ ID NO:19), Stearatoyl-Ser-Leu-Ile-Gly-Lys-NH_2 (SEQ ID NO:20), Stearatoyl-Leu-Ile-Gly-Arg-OH (SEQ ID NO:21), Stearatoyl-Leu-Ile-Gly-Arg-Leu-OH (SEQ ID NO:22), Stearatoyl-Leu-Ile-Gly-Lys-OH (SEQ ID NO:23), Stearatoyl-Ser-Leu-Ile-Gly-Lys-OH (SEQ ID NO:24), H_2-Ser-Leu-Ile-Gly-Arg-Leu-NH_2 (SEQ.ID.No.25), H_2-Ser-Leu-Ile-Gly-Arg-Leu-OH (SEQ.ID.No.26), Palmitoyl-Ser-Leu-Ile-Gly-Arg-Leu-NH_2 (SEQ.ID.No.27), Palmitoyl-Ser-Leu-Ile-Gly-Arg-Leu-OH (SEQ.ID.No.28), Stearatoyl-Ser-Leu-Ile-Gly-Arg-Leu-NH_2 (SEQ.ID.No.29), and Stearatoyl-Ser-Leu-Ile-Gly-Arg-Leu-OH (SEQ.ID.No.30), or a cosmetically-acceptable salt thereof.

The symbol A_1, A_2, or the like used herein (e.g., in Figure 1) stands for the residue of an alpha-amino acid. Such symbols represent the general structure, -NH-CH(X)-CO- or =N-CH(X)-CO- when it is at the N-terminus or -NH-CH(X)-CO- when it is not at the N-terminus, where X denotes the side chain (or identifying group) of the alpha-amino acid, e.g., X is $-CH(CH_3)_2$ for Val. Note that the N-terminus is at the left and the C-terminus at the right in accordance with the conventional representation of a polypeptide chain. R_1 and R_2 are both bound to the free nitrogen atom N-terminal amino acid (e.g., A_1 or A_2) and the R_3 is bound

to the free carboxy group of the C-terminal amino acid (e.g., A_5, A_6, or A_7).

"Cha" herein refers to cyclohexylalanine, "2,3-diaP" refers to 2,3-diaminoproprionic acid, and "Har" refers to homoarginine. Furthermore, where the amino acid residue is optically active, it is the L-form configuration that is intended unless the D-form is expressly designated. An alkyl group, if not specified, contains 1-12 carbon atoms.

The peptide of the invention can be provided in the form of cosmetically acceptable salts. Examples of preferred salts are those with therapeutically acceptable organic acids, e.g., acetic, palmitic, oleic, stearic, lactic, maleic, citric, malic, ascorbic, succinic, benzoic, salicylic, methanesulfonic, or pamoic acid, as well as polymeric acids such as tannic acid or carboxymethyl cellulose, and salts with inorganic acids such as the hydrohalic acids (e.g., hydrochloric acid), sulfuric acid or phosphoric acid.

The amount of peptide present in the composition will depend on the peptide used. The peptide typically will be present in the composition in an amount from about 0.001% to about 10% by weight, in particular in an amount from about 0.005% to about 5% by weight.

The method for synthesizing peptides of the present invention are well documented and are within the ability of a person of ordinary skill in the art. See, e.g., Bodanszky M, Int J Pept Protein Res

25(5):449-74 (1985), Fmoc Solid Phase Peptide Synthesis, eds. Chan, W. & White, P. (Oxford University Press, 2000), and Chemial Approaches to the Synthesis of Peptides and Proteins, Lloyd-Williams, P. et al. (CRC Press, 1997).

Topical Compositions

The topical compositions useful in the present invention involve formulations suitable for topical application to skin. In one embodiment, the composition comprises the He Shou Wu extract and a cosmetically-acceptable topical carrier. In one embodiment, the cosmetically-acceptable topical carrier is from about 50% to abut 99.99%, by weight, of the composition (e.g., from about 80% to about 95%, by weight, of the composition.

The compositions may be made into a wide variety of product types that include but are not limited to lotions, creams, gels, sticks, sprays, ointments, cleansing liquid washes and solid bars, pastes, foams, powders, mousses, shaving creams, wipes, patches, nail lacquers, wound dressing and adhesive bandages, hydrogels, films and make-up such as foundations, mascaras, and lipsticks. These product types may comprise several types of cosmetically- acceptable topical carriers including, but not limited to solutions, emulsions (e.g., microemulsions and nanoemulsions), gels, solids and liposomes. The following are non-limitative examples of such

carriers. Other carriers can be formulated by those of ordinary skill in the art.

The topical compositions useful in the present invention can be formulated as solutions. Solutions typically include an aqueous or organic solvent (e.g., from about 50% to about 99.99% or from about 90% to about 99% of a cosmetically acceptable aqueous or organic solvent). Examples of suitable organic solvents include: propylene glycol, polyethylene glycol (200-600), polypropylene glycol (425-2025), glycerol, 1,2,4-butanetriol, sorbitol esters, 1,2,6-hexanetriol, ethanol, and mixtures thereof.

Topical compositions useful in the subject invention may be formulated as a solution comprising an emollient. Such compositions preferably contain from about 2% to about 50% of an emollient(s). As used herein, "emollients" refer to materials used for the prevention or relief of dryness, as well as for the protection of the skin. Examples of emollients include, but are not limited to, those set forth in the International Cosmetic Ingredient Dictionary and Handbook, eds. Wenninger and McEwen, pp. 1656-61, 1626, and 1654-55 (The Cosmetic, Toiletry, and Fragrance Assoc., Washington, D.C., 7th Edition, 1997) (hereinafter "ICI Handbook").

A lotion can be made from such a solution. Lotions typically comprise from about 1% to about 20% (e.g., from about 5% to about 10%) of an emollient(s) and from about 50% to about 90% (e.g., from about 60% to about 80%) of water.

Another type of product that may be formulated from a solution is a cream. A cream typically comprises from about 5% to about 50% (e.g., from about 10% to about 20%) of an emollient(s) and from about 45% to about 85% (e.g., from about 50% to about 75%) of water.

Yet another type of product that may be formulated from a solution is an ointment. An ointment may comprise a simple base of animal or vegetable oils or semi-solid hydrocarbons. An ointment may comprise from about 2% to about 10% of an emollient(s) plus from about 0.1% to about 2% of a thickening agent(s). Examples of thickening agents include, but are not limited to, those set forth in the ICI Handbook pp. 1693-1697.

The topical compositions useful in the present invention formulated as emulsions. If the carrier is an emulsion, from about 1% to about 10% (e.g., from about 2% to about 5%) of the carrier comprises an emulsifier(s). Emulsifiers may be nonionic, anionic or cationic. Examples of emulsifiers include, but are not limited to, those set forth in the ICI Handbook, pp.1673-1686.

Lotions and creams can be formulated as emulsions. Typically such lotions comprise from 0.5% to about 5% of an emulsifier(s). Such creams would typically comprise from about 1% to about 20% (e.g., from about 5% to about 10%) of an emollient(s); from about 20% to about 80% (e.g., from 30% to about 70%)

of water; and from about 1% to about 10% (e.g., from about 2% to about 5%) of an emulsifier(s).

Single emulsion skin care preparations, such as lotions and creams, of the oil-in-water type and water-in-oil type are well-known in the cosmetic art and are useful in the subject invention. Multiphase emulsion compositions, such as the water-in-oil-in-water type are also useful in the subject invention. In general, such single or multiphase emulsions contain water, emollients, and emulsifiers as essential ingredients.

The topical compositions of this invention can also be formulated as a gel (e.g., an aqueous, alcohol, alcohol/water, or oil gel using a suitable gelling agent(s)). Suitable gelling agents for aqueous and/or alcoholic gels include, but are not limited to, natural gums, acrylic acid and acrylate polymers and copolymers, and cellulose derivatives (e.g., hydroxymethyl cellulose and hydroxypropyl cellulose). Suitable gelling agents for oils (such as mineral oil) include, but are not limited to, hydrogenated butylene/ethylene/styrene copolymer and hydrogenated ethylene/propylene/styrene copolymer. Such gels typically comprises between about 0.1% and 5%, by weight, of such gelling agents.

The topical compositions of the present invention can also be formulated into a solid formulation (e.g., a wax-based stick, soap bar composition, powder, or a wipe containing powder).

Liposomal formulations are also useful compositions of the subject invention. In one embodiment, the He Shou Wu extract and/or the pigment and/or the peptide of formula I are contained within the liposome. Examples of liposomes are unilamellar, multilamellar, and paucilamellar liposomes, which may or may not contain phospholipids. Such compositions can be prepared by first combining hesperetin with a phospholipid, such as dipalmitoylphosphatidyl choline, cholesterol and water. Epidermal lipids of suitable composition for forming liposomes may be substituted for the phospholipid. The liposome preparation may then incorporated into one of the above carriers (e.g., a gel or an oil-in-water emulsion) in order to produce the liposomal formulation.

In one-embodiment, the liposome is non-ionic. In one example, the liposome contains (a) glycerol dilaurate; (b) compounds having the steroid backbone found in cholesterol; and (c) fatty acid ethers having from about 12 to about 18 carbon atoms. In a further embodiment, the liposome comprises glycerol dilaurate, cholesterol, polyoxyethylene-10-stearyl ether, and polyoxyethylene-9-lauryl ether. In one embodiment, these ingredients are in a ratio of about 38:12:33:17.

In one embodiment, the liposomes are present in the topical composition in an amount, based upon the total volume of the composition, of from about 5 mg/ml to about 100 mg/ml such as from about 10 mg/ml to about 50 mg/ml. Methods of preparing liposomes are well

known in the art, such as those disclosed in U.S. Patent No. 5,013,497 and 5,260,065.

The topical compositions useful in the subject invention may contain, in addition to the aforementioned components, a wide variety of additional oil-soluble materials and/or water-soluble materials conventionally used in compositions for use on skin, hair, and nails at their art-established levels.

Additional Cosmetically Active Agents

In one embodiment, the topical composition further comprises another cosmetically active agent in addition to the He Shou Wu extract and pigments. What is meant by a "cosmetically active agent" is a compound (e.g., a synthetic compound or a compound isolated from a natural source) that has a cosmetic or therapeutic effect on the skin, hair, or nails, including, but not limiting to, lightening agents, darkening agents such as self-tanning agents, anti-acne agents, shine control agents, anti-microbial agents, anti-inflammatory agents, anti-mycotic agents, anti-parasite agents, external analgesics, sunscreens, photoprotectors, antioxidants, keratolytic agents, detergents/surfactants, moisturizers, nutrients, vitamins, energy enhancers, anti-perspiration agents, astringents, deodorants, hair removers, firming agents, anti-callous agents, and agents for hair, nail, and/or skin conditioning.

In one embodiment, the agent is selected from, but not limited to, the group consisting of hydroxy acids, benzoyl peroxide, D-panthenol, octyl methoxycinnimate, titanium dioxide, octyl salicylate, homosalate, avobenzone, carotenoids, free radical scavengers, spin traps, retinoids such as retinol and retinyl palmitate, ceramides, polyunsaturated fatty acids, essential fatty acids, enzymes, enzyme inhibitors, minerals, hormones such as estrogens, steroids such as hydrocortisone, 2-dimethylaminoethanol, copper salts such as copper chloride, peptides containing copper such as Cu:Gly-His-Lys, coenzyme Q10, peptides such as those disclosed in PCT Patent Application No. WO 00/15188, amino acids such a proline and tyrosine, vitamins, lactobionic acid, acetyl-coenzyme A, niacin, riboflavin, thiamin, ribose, electron transporters such as NADH and FADH2, and other botanical extracts such as aloe vera, and derivatives and mixtures thereof. The cosmetically active agent will typically be present in the composition of the invention in an amount of from about 0.001% to about 20% by weight of the composition, e.g., about 0.005% to about 10% such as about 0.01% to about 5%.

Examples of vitamins include, but are not limited to, vitamin A, vitamin Bs such as vitamin B3, vitamin B5, and vitamin B12, vitamin C, vitamin K, and vitamin E and derivatives thereof.

Examples of hydroxy acids include, but are not limited, to glycolic acid, lactic acid, malic acid,

salicylic acid, citric acid, and tartaric acid. See, e.g., European Patent Application No. 273,202.

Examples of antioxidants include, but are not limited to, water-soluble antioxidants such as sulfhydryl compounds and their derivatives (e.g., sodium metabisulfite and N-acetyl-cysteine), lipoic acid and dihydrolipoic acid, resveratrol, lactoferrin, and ascorbic acid and ascorbic acid derivatives (e.g., ascorbyl palmitate and ascorbyl polypeptide). Oil-soluble antioxidants suitable for use in the compositions of this invention include, but are not limited to, butylated hydroxytoluene, retinoids (e.g., retinol and retinyl palmitate), tocopherols (e.g., tocopherol acetate), tocotrienols, and ubiquinone. Natural extracts containing antioxidants suitable for use in the compositions of this invention, include, but not limited to, extracts containing flavonoids and isoflavonoids and their derivatives (e.g., genistein and diadzein), extracts containing resveratrol and the like. Examples of such natural extracts include grape seed, green tea, pine bark, and propolis.

Other Materials

Various other materials may also be present in the compositions useful in the subject invention. These include humectants, proteins and polypeptides, preservatives and an alkaline agent. Examples of such agents are disclosed in the ICI Handbook, pp.1650-1667.

The compositions of the present invention may also comprise chelating agents (e.g., EDTA) and preservatives (e.g., parabens). Examples of suitable preservatives and chelating agents are listed in pp. 1626 and 1654-55 of the ICI Handbook. In addition, the topical compositions useful herein can contain conventional cosmetic adjuvants, such as dyes, opacifiers (e.g., titanium dioxide), pigments, and fragrances.

Mineral Water

The compositions of the present invention may be prepared using a mineral water, for example mineral water that has been naturally mineralized such as Evian® Mineral Water (Evian, France). In one embodiment, the mineral water has a mineralization of at least about 200 mg/L (e.g., from about 300 mg/L to about 1000 mg/L). In one embodiment, the mineral water comprises at least about 10 mg/L of calcium and/or at least about 5 mg/L of magnesium.

The composition and formulations containing such compositions of the present invention may be prepared using methodology that is well known by an artisan of ordinary skill.

Example 1: He Shou Wu Induces Pigmentation in Culture

He Shou Wu was tested for its effect on pigmentation, using keratinocyte-melanocyte cultures, DOPA staining and computerized image analysis. The He

Shou Wu extract was a solid obtained from an aqueous extract from the plant's root (He Shou Wu, JiangYing TianJiang Pharmaceutical, Inc., China). The extract was dissolved in culture medium and was assayed at a 0.001% (w/v) concentration.

Human HaCaT keratinocytes (as described in: Boukamp P., et al., J Cell Biol 106, 3, 761-771, 1988) were maintained in Dulbecco's Modified Eagle's Medium (DMEM) containing 10% fetal bovine serum (FBS), 4.5 mg/ml glucose, 2 mM L-glutamine, 50 U/ml penicillin and 50 □g /ml streptomycin (Life Technologies, Gaithersburg, MD). Cells were maintained at <80% confluency in 5% CO_2 (v/v) and were used in experimental procedures up to culture passage 15. Human primary melanocytes (Clonetics, San Diego, CA or Cascade Biologics, Portland, OR) were maintained according to manufacturer's instructions. To establish keratinocyte-melanocyte co-cultures, $6x10^4$ melanocytes were plated in each well of a 24 well plate and maintained according to manufacturer's instructions. Melanocytes were rinsed three times with melanocyte growth media without PMA, and keratinocytes ($6x10^4$) were plated to establish the co-culture. Co-cultures were treated for three days with He Shou Wu and assayed for cell viability and pigment level on the forth day. Cell viability was assayed using alamarBlue™ (Acumed International, West Lake, OH) following manufacturer's instructions. No change in viability was observed following three daily

treatments with 0.001% of He Shou Wu. All *in vitro* experiments were performed in triplicates.

Following three daily treatments, the co-cultures were briefly fixed (10% buffered formalin from Fisher Scientific, 15 minutes), washed three times with Phosphate-buffered saline (PBS, from Life Technologies) and stained with L-3,4-Dihydroxyphenylalanine (DOPA, from Sigma, St. Louis, MO) 0.1% in PBS, for 5 hours at 37°C, followed by two PBS washes and formalin (10%) fixation. DOPA is a substrate for tyrosinase, therefore an increase in staining represents increased tyrosinase activity and pigment production. DOPA-stained cells were used for image analysis. All images were obtained and analyzed with Image Pro Plus 4.0 software (Media Cybernetics, Silver Spring, MD). Parameters measured were surface area of stained material within melanocyte and keratinocytes and the total surface area of the cells in the culture, and the relative pigmented area was calculated. A value of 100% was assigned to untreated controls, and values of treatment groups were normalized to their relevant controls. In all experiments there was no difference between PBS-treated cells and untreated controls. Data are presented with standard deviation (SigmaPlot® 5.0, SPSS Science, Chicago, IL).

Table 1 shows the results of representative co-culture experiments, normalized for their relative controls, demonstrating that He Shou Wu treatment enhanced pigmentation. This Table demonstrates the specificity of the compositions of this invention in

inducing pigmentation (e.g., increasing pigmentation by up to 218%).

TABLE 1

Test Material	Conc.	% increase in pigmentation
Control	-	-
He Shou Wu	0.001%(W/V)	218

Example 2: He Shou Wu Induces Pigmentation in Pigmented Epidermal Equivalents

He Shou Wu extract of Example 1 was also tested for its ability to induce pigmentation in pigmented epidermal equivalents. The pigmented epidermal equivalents contain human normal melanocytes, together with normal, human-derived epidermal keratinocytes, which have been cultured to form a multi-layered, highly differentiated model of the human epidermis. Type II pigmented epidermal equivalents (consists of normal human keratinocytes pooled from variety of phototype skins and normal human melanocytes derived from type II phototype skin) were treated with test compounds for three or five days and samples were harvested on the fourth or sixth day of the study. The harvested equivalents were stained with DOPA (a substrate for tyrosinase) or with Fontana-Mason (F&M)

(Sheenan DC, Hrapckak BB, eds: Theory and practice of Histo-Thchnology (St Louis: CV Mosby, 1980) pp 223-277). F&M staining identifies silver nitrate reducing activity, which, in skin, identifies melanin.

The Epidermal equivalents used were SkinEthic® reconstructed human epidermis from SkinEthic™ Laboratory (Nice, France). UV irradiation was performed with a UVB FS light source in an exposure chamber, with plate covers removed and Phosphate-buffered saline (PBS, from Gibco-BRL, Gaithersburg, MD) present in the lower chamber. UVB intensity was measured with a UVX radiometer (UVP Inc., San Gabriel, CA). Equivalents were treated with 0.1-0.12 J/cm2. No loss of viability was observed in equivalents treated with up to 0.3 J/cm2. He Shou Wu extract was assayed at 0.01-0.1% (w/v) concentration, and was dissolved in PBS.

On the fourth or sixth day of the study, the equivalents were fixed, sectioned and F&M stained, or they were DOPA stained as whole without sectioning, using standard techniques. Images were captured as described in Example 1. At least three sections per equivalent, three equivalents per experiment were processed. Each experiment was repeated three times. DOPA-stained epidermal equivalents were evaluated for the change in tyrosinase activity. F&M-stained histological sections were evaluated for the change in pigment deposition. Due to the low content of melanin within the type II epidermal equivalent, it was not possible to quantify the level of pigment within

melanocytes in F&M stained sections by image analysis. Therefore, we evaluate the pigment change using the scale defined in Table 2.

TABLE 2

Score	Description
0	No change in DOPA staining and in melanin deposition
1	Minimal increase in DOPA staining and/ or in pigment deposition
2	Increased DOPA staining and/ or in pigment deposition
3	Strong increase in DOPA staining and/ or in pigment deposition

Table 3 represents the overall score in change of pigmentation, as evaluated by DOPA and F&M staining, as set forth above, when equivalents were exposed to He Shou Wu (0.01% and 0.1% (w/v)), and UVB irradiation (0.10J/cm^2). This Table demonstrates that He Shou Wu treatment resulted in darkening levels similar to those produced by UVB irradiation.

TABLE 3

Test Material	Score		Overall Score
	DOPA staining (tyrosinase activity)	F&M staining (Pigment deposition)	

Control	0	0	0
UVB (positive control)	3	2-3	2-3
He Shou Wu 0.01%	2-3	1-2	1-2
He Shou Wu 0.1%	2-3	3	2-3

Example 3: He Shou Wu Induces Pigmentation In Vivo

Dark skinned Yucatan microswine (Charles River, Portland, ME) were housed in appropriately sized cages in an environmentally controlled room with a 12-hour light - 12-hour dark photoperiod and supplied with food and water *ad libitum*. Twenty µl of test materials were applied topically, twice a day, five days/week, for eight or nine weeks, on the dorsum of the swine. Treatments of individual swine were always arranged in a head to tail order on one side, and in a tail to head order on the other side of the animal. Biopsies were taken using standard techniques. All swine studies presented here had no visual irritation, and histological analyses revealed no markers of irritation or other pathological signs.

Swine were treated with either 1% (w/v) of the He Shou Wu extract of Example 1 or ultraviolet-B radiation (as a positive control). The He Shou Wu extract was dissolved in ethanol: propylene glycol 70:30 (v/v). A mean erythema dose (MED) of UVB was determined by placing a plastic template with 1x1 $inch^2$ cutouts on the dorsum of the swine. Using a UVB lamp (Model UVM-57, 302nm lamp, UVP Inc., Upland, CA)

placed on the template, sites were exposed to UVB with increasing time points, every other day for five days. Unexposed sites were covered with the same material as the template. One MED was established as the dose that produces the least amount of visible erythema. Swine were exposed to one MED, once per day, on three alternate days (Mon, Wed, Fri). All swine studies presented here had no visual irritation, and histological analyses revealed no markers of irritation or other pathological signs.

Following eight weeks of treatment, skin biopsies were taken using standard methods, for pigment deposition analysis. Sections from the skin biopsies were stained with Hematoxylin and Eosin (H&E), or with Fontana-Mason (F&M), using standard procedures (Sheenan DC, Hrapckak BB, eds., Theory and Practice of Histo-Technology (The C. V. Mosby Co., St. Louis (1980) pp. 223-277). At least three sections per biopsy were processed. Each experiment was repeated at least two times.

Histological analysis revealed an increase in pigment deposition in swine treated with He Shou Wu. Criteria for evaluation were total increase in pigment deposition, and the presence of capped epidermal cells above the basal layer. Table 5 represents the average value of all sites of responsive swine treated with each test material. The scale for evaluation is defined in Table 4.

TABLE 4

Score	Description
-1	Slight lightening
0	No change
1	Minimal increase in pigment deposition
2	Increased pigment deposition
3	Strong increase in pigment deposition, some increase in caps
4	Strong increase in pigment deposition, strong increase in caps

TABLE 5

Compositions	Score
Control	0
Ethanol: polypropylene glycol	0
UVB	4
1% (w/v) He Shou Wu	2-3

This example demonstrates that He Shou Wu enhanced pigment deposition in, and thereby darkened, live skin.

It is understood that while the invention has been described in conjunction with the detailed description thereof, that the foregoing description is intended to illustrate and not limit the scope of the invention, which is defined by the scope of the appended claims. Other aspects, advantages, and modifications are within the claims.

What is claimed is:

Claims

1. A composition for darkening the skin, comprising a safe and effective amount of a He Shou Wu extract and a cosmetically-acceptable carrier.

2. A composition of claim 1, wherein said composition further comprises a pigment.

3. A composition of claim 2, wherein said pigment is melanin, a derivative of melanin, $CuCl_2$, Hedychium extract, or Bearberry extract.

4. A composition of claim 1, wherein said composition further comprises a peptide of the formula

$$R_1$$
$$>A_1\text{-}A_2\text{-}A_3\text{-}A_4\text{-}A_5\text{-}A_6\text{-}A_7\text{-}R_3$$
$$R_2$$

wherein:

A_1 is Ser or 2,3-diaP, or is absent;
A_2 is Val, Leu, Ile, or Cha;
A_3 is Val, Leu, Ile, or Cha;
A_4 is Gly or Ala;
A_5 is Lys, Arg, or Har;
A_6 is Val, Leu, Ile, or Cha, or is absent;

A_7 is Asp or Glu, or is absent; provided, A_7 is absent if A_6 is absent;

each R_1 and R_2, independently, is H, C_{1-12} alkyl, C_{7-10} phenylalkyl, or $C(=O)E_1$, where E_1 is C_{1-20} alkyl, C_{3-20} alkenyl, C_{3-20} alkynyl, phenyl, 3,4-dihydroxyphenylalkyl, naphthyl, or C_{7-10} phenylalkyl; provided that when either R_1 or R_2 is $C(=O)E_1$, the other must be H; and

R_3 is OH, NH_2, C_{1-12} alkoxy, C_{7-10} phenylalkoxy, C_{11-20} naphthylalkoxy, C_{1-12} alkylamino, C_{7-10} phenylalkylamino, or C_{11-20} naphthylalkylamino; or a cosmetically acceptable salt thereof.

5. A composition of claim 1, wherein said composition comprises from about 0.001%, by weight, to about 20%, by weight, of said He Shou Wu extract and said cosmetically-acceptable carrier is a topical carrier.

6. A composition of claim 2, wherein said composition comprises from about 0.001%, by weight, to about 20%, by weight, of said He Shou Wu extract, from about 0.001%, by weight, to about 20%, by weight, of said pigment, and said cosmetically-acceptable carrier is a topical carrier.

7. A composition of claim 3, wherein said composition comprises from about 0.001%, by weight, to about 20%, by weight, of said He Show Wu extract, from about 0.001%, by weight, to about 20%, by weight, of

said pigment, and said cosmetically-acceptable carrier is a topical carrier.

8. A composition of claim 4, wherein said composition comprises from about 0.001%, by weight, to about 20%, by weight, of said He Shou Wu extract, from about 0.001%, by weight, to about 20%, by weight, of said peptide, and said cosmetically-acceptable carrier is a topical carrier.

9. A method of darkening the skin, said method comprising topically applying to the skin a composition comprising a safe and effective amount of a He Shou Wu extract.

10. A method of claim 9, wherein said composition further comprises a pigment.

11. A method of claim 10, wherein said pigment is melanin, a derivative of melanin, $CuCl_2$, Hedychium extract, or Bearberry extract.

12. A method of claim 9, wherein said composition further comprises a peptide of the formula

$$R_1$$
$$>A_1\text{-}A_2\text{-}A_3\text{-}A_4\text{-}A_5\text{-}A_6\text{-}A_7\text{-}R_3$$
$$R_2$$

wherein:

A_1 is Ser or 2,3-diaP, or is absent;

A_2 is Val, Leu, Ile, or Cha;

A_3 is Val, Leu, Ile, or Cha;

A_4 is Gly or Ala;

A_5 is Lys, Arg, or Har;

A_6 is Val, Leu, Ile, or Cha, or is absent;

A_7 is Asp or Glu, or is absent; provided, A_7 is absent if A_6 is absent;

each R_1 and R_2, independently, is H, C_{1-12} alkyl, C_{7-10} phenylalkyl, or $C(=O)E_1$, where E_1 is C_{1-20} alkyl, C_{3-20} alkenyl, C_{3-20} alkynyl, phenyl, 3,4-dihydroxyphenylalkyl, naphthyl, or C_{7-10} phenylalkyl; provided that when either R_1 or R_2 is $C(=O)E_1$, the other must be H; and

R_3 is OH, NH_2, C_{1-12} alkoxy, C_{7-10} phenylalkoxy, C_{11-20} naphthylalkoxy, C_{1-12} alkylamino, C_{7-10} phenylalkylamino, or C_{11-20} naphthylalkylamino; or a cosmetically acceptable salt thereof.

13. A method of claim 9, wherein said composition comprises from about 0.001%, by weight, to about 20%, by weight, of said He Shou Wu extract and said composition comprises a cosmetically-acceptable topical carrier.

14. A method of claim 10, wherein said composition comprises from about 0.001%, by weight, to about 20%, by weight, of said He Shou Wu extract and said composition comprises a cosmetically-acceptable topical carrier.

15. A method of claim 11, wherein said composition comprises from about 0.001%, by weight, to about 20%, by weight, of said He Shou Wu extract and said composition comprises a cosmetically-acceptable topical carrier.

16. A method of claim 12, wherein said composition comprises from about 0.001%, by weight, to about 20%, by weight, of said He Shou Wu extract and said composition comprises a cosmetically-acceptable topical carrier.

17. A product comprising:

(a) a composition for darkening the skin, wherein said composition comprises a safe and effective amount of a He Shou Wu extract; and

(b) instructions directing the user to apply said composition to the skin to darken the skin.

18. A product of claim 17, wherein said composition further comprises a pigment.

19. A product of claim 18, wherein said pigment is melanin, a derivative of melanin, $CuCl_2$, Hedychium extract, or Bearberry extract.

20. A product of claim 17, wherein said composition further comprises a peptide of the formula

R_1

$>A_1\text{-}A_2\text{-}A_3\text{-}A_4\text{-}A_5\text{-}A_6\text{-}A_7\text{-}R_3$

R_2

wherein:

A_1 is Ser or 2,3-diaP, or is absent;

A_2 is Val, Leu, Ile, or Cha;

A_3 is Val, Leu, Ile, or Cha;

A_4 is Gly or Ala;

A_5 is Lys, Arg, or Har;

A_6 is Val, Leu, Ile, or Cha, or is absent;

A_7 is Asp or Glu, or is absent; provided, A_7 is absent if A_6 is absent;

each R_1 and R_2, independently, is H, $C_{1\text{-}12}$ alkyl, $C_{7\text{-}10}$ phenylalkyl, or $C(=O)E_1$, where E_1 is $C_{1\text{-}20}$ alkyl, $C_{3\text{-}20}$ alkenyl, $C_{3\text{-}20}$ alkynyl, phenyl, 3,4-dihydroxyphenylalkyl, naphthyl, or $C_{7\text{-}10}$ phenylalkyl; provided that when either R_1 or R_2 is $C(=O)E_1$, the other must be H; and

R_3 is OH, NH_2, $C_{1\text{-}12}$ alkoxy, $C_{7\text{-}10}$ phenylalkoxy, $C_{11\text{-}20}$ naphthylalkoxy, $C_{1\text{-}12}$ alkylamino, $C_{7\text{-}10}$ phenylalkylamino, or $C_{11\text{-}20}$ naphthylalkylamino; or a cosmetically acceptable salt thereof.

21. A product of claim 17, wherein said composition comprises from about 0.001%, by weight, to about 20%, by weight, of said He Shou Wu extract and said composition comprises a cosmetically-acceptable topical carrier.

22. A product of claim 18, wherein said composition comprises from about 0.001%, by weight, to about 20%, by weight, of said He Shou Wu extract and said composition comprises a cosmetically-acceptable topical carrier.

23. A product of claim 19, wherein said composition comprises from about 0.001%, by weight, to about 20%, by weight, of said He Shou Wu extract and said composition comprises a cosmetically-acceptable topical carrier.

24. A product of claim 20, wherein said composition comprises from about 0.001%, by weight, to about 20%, by weight, of said He Shou Wu extract and said composition comprises a cosmetically-acceptable topical carrier.

25. A method of promoting a product comprising a composition where said composition comprises a safe and effective amount of a He Shou Wu, wherein said method comprises directing the user to apply said composition to the skin to darken the skin.

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C. DOCUMENTS CONSIDERED TO BE RELEVANT

Category *	Citation of document, with indication, where appropriate, of the relevant passages	Relevant to claim No.
X	DATABASE WPI Week 199831 Derwent Publications Ltd., London, GB; AN 1998-357420 XP002252584 & JP 10 139677 A (OMORI Y), 26 May 1998 (1998-05-26) abstract	1,2,5,6
Y		17,18, 21,22
Y	WO 01 68048 A (UNILEVER PLC ET AL.) 20 September 2001 (2001-09-20) claims 8,9	17,18, 21,22
	-/--	

[X] Further documents are listed in the continuation of box C.

[X] Patent family members are listed in annex.

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C.(Continuation) DOCUMENTS CONSIDERED TO BE RELEVANT

Category *	Citation of document, with indication, where appropriate, of the relevant passages	Relevant to claim No.
X	DATABASE WPI Week 198831 Derwent Publications Ltd., London, GB; AN 1988-217499 XP002252585 & JP 63 154604 A (EIKODO KK), 27 June 1988 (1988-06-27) abstract	1,5
X	DATABASE WPI Week 199549 Derwent Publications Ltd., London, GB; AN 1995-379970 XP002252586 & JP 07 258063 A (KOSE KK), 9 October 1995 (1995-10-09) abstract	1,5
X	GB 2 240 715 A (MICHAEL FUK YAU WAN) 14 August 1991 (1991-08-14) claims 1,14-16 page 5, paragraph 2	1,5

INTERNATIONAL SEARCH REPORT

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Patent document cited in search report	Publication date	Patent family member(s)	Publication date
JP 10139677 A	26-05-1998	NONE	
WO 0168048 A	20-09-2001	AU 6210801 A WO 0168048 A2	24-09-2001 20-09-2001
JP 63154604 A	27-06-1988	NONE	
JP 7258063 A	09-10-1995	NONE	
GB 2240715 A	14-08-1991	NONE	

Form PCT/ISA/210 (patent family annex) (July 1992)